

## Histone H2A.XS139ph antibody (pAb)

**Catalog Nos:** 39117, 39118

**RRID:** AB\_2793161

**Application(s):** DB, ICC, IF, WB

**Reactivity:** Human, Wide Range Predicted

**Quantities:** 100 µg, 10 µg

**Purification:** Protein A Chromatography

**Host:** Rabbit

**Isotype:** IgG

**Molecular Weight:** 15 kDa

**Background:** Histone H2AX phospho Ser139 (H2AX, H2A histone family member X) replaces conventional H2A in a subset of nucleosomes. Nucleosomes wrap and compact DNA into chromatin, limiting DNA accessibility to the cellular machineries that require DNA as a template. Histones thereby play a central role in transcriptional regulation, DNA repair, DNA replication and chromosomal stability. DNA accessibility is regulated via a complex set of post-translational modifications of histones, also called the histone code, and nucleosome remodeling. Histone H2AX is required for checkpoint-mediated arrest of cell cycle progression in response to low doses of ionizing radiation, and for efficient repair of DNA double-strand breaks (DSBs), specifically when modified by C-terminal phosphorylation.

**Immunogen:** This Histone H2AX phospho Ser139 antibody was raised against a peptide including phosphoserine 139 of histone H2AX.

**Buffer:** PBS containing 30% glycerol, 0.035% sodium azide. Sodium azide is highly toxic.

### Application Notes:

Applications Validated by Active Motif:

ICC/IF: 1:500 dilution

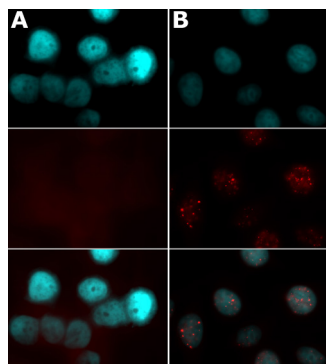
WB\*: 1:1,000 - 1:5,000 dilution

DB: 0.1 µg/ml

\*Note: many chromatin-bound proteins are not soluble in a low salt nuclear extract and fractionate to the pellet. Therefore, we recommend a High Salt / Sonication Protocol when preparing nuclear extracts for Western Blot. Visit [activemotif.com](http://activemotif.com) to download the protocol.

**Storage and Guarantee:** Some products may be shipped at room temperature. This will not affect their stability or performance. Avoid repeated freeze/thaw cycles by aliquoting items into single-use fractions for storage at -20°C for up to 2 years. Keep all reagents on ice when not in storage. This product is guaranteed for 12 months from date of receipt.

This product is for research use only and is not for use in diagnostic procedures.



#### Histone H2AX phospho Ser139 antibody tested by immunofluorescence.

HeLa cells stained with Histone H2AX phospho Ser139 antibody (1:500 dilution) using MAX Stain™ Immunofluorescence Tools. The HeLa cells were blocked with MAXblock™ Blocking Medium and stained with Histone H2AX phospho Ser139 antibody.

Panel A: Untreated HeLa cells.

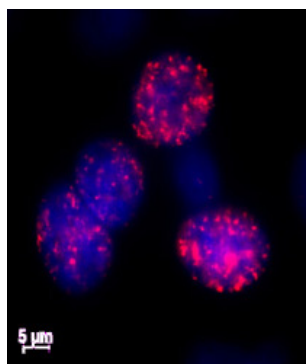
Panel B: Cells fixed and stained 90 minutes after 3 Gy ionizing radiation treatment.

Top images: Cells were stained with DAPI.

Middle images: Same cells stained with Histone H2AX phospho Ser139 antibody.

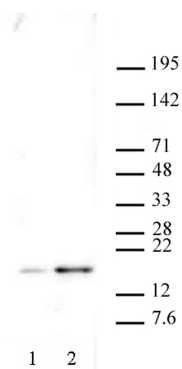
Bottom images: Merge of both images above.

Images were made using Zeiss Axiovision with equivalent acquisition settings for direct comparison. Note Panel B-middle image, which shows intense nuclear clustering of ionizing radiation-induced phosphorylation of Ser139 of H2AX. In contrast, Panel A-middle image shows



#### Histone H2AX phospho Ser139 antibody tested by immunofluorescence.

Etoposide-treated HeLa cells stained with Cat. No. 39117 (1:500 dilution, red) and counterstained with DAPI (blue).

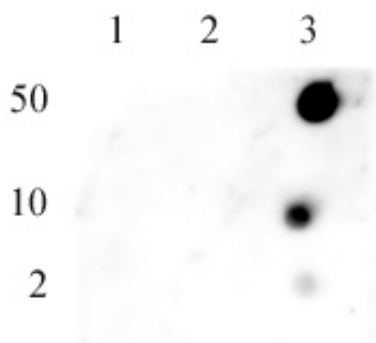


#### Histone H2AX phospho Ser139 antibody tested by Western blot.

Western blot: Nuclear extract of U2OS cells (20 µg per lane) probed with Histone H2AX phospho Ser139 polyclonal antibody (1:500 dilution).

Lane 1: untreated cells

Lane 2: cells treated with camptothecin



#### Dot blot analysis was used to confirm the specificity of Histone H2A.XS139ph

Peptides were spotted onto nitrocellulose at 2, 10 and 50 picomoles and probed with the antibody at 0.1 µg/mL. Col. 1: Unmodified Peptide of Histone H2AX. Col. 2: Peptide containing phospho-tyrosine 142 of Histone H2AX. Col. 3: Peptide containing phospho-serine 139 of Histone H2AX.